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ORIGINAL PAPER

Variability and divergence in *Pongamia pinnata* for further use in tree improvement

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Abstract: A total of 24 candidate plus trees (CPTs) of *Pongamia pinnata* (L.) Pierre. were selected to elucidate their variation and diversity based on thirteen quantitative traits (4 pod traits, 6 seed traits of parent trees and 3 progeny traits) at Forest Research Centre, Institute of Forest Productivity - Mandar, Ranchi district during 2005–2007. The results show that, CPT-19 had maximum for seven traits viz, pod length (65.6 mm), 100-pod weight (542.4 g), seed 2D (two dimension) area (351.2 mm^2), seed length (27.9 mm), seed breadth (17.4 mm), 100-seed weight (217.9 g) and plant height (164.3 cm). The traits, 100-pod weight and 100-seed weight had a high heritability (98.4%, 96.9%) accompanied with high genetic advance (46.0%, 34.9%). There is a positive significant correlation between 100-pod weight and 100-seed weight traits at both genotypic and phenotypic levels with plant height, collar diameter and volume index at 30 MAS (months after sowing). Volume index expressed a moderate heritability (47.4%) accompanied with high genetic advance (48.4%), indicating that the character is governed by additive gene effects. In divergence study, 24 accessions were grouped into 6 clusters on the basis of non-hierarchical euclidian cluster analysis. The genotypes in cluster IV (CPT-5, CPT-6, CPT-7, CPT-12, CPT-16, CPT-18, CPT-22) and cluster III (CPT-4, CPT-8, CPT-9, CPT-20, CPT-21) were most heterogeneous and can be best used within group hybridization. The wide diversity exists between the cluster V and II, followed by cluster II and I and crosses between CPTs of these clusters may result in substantial segregates. It is revealed that the existence of substantial variation and diversity can be utilized for genetic resource conservation and further tree improvement programmers of the species.

Keywords: *Pongamia pinnata*; heritability; genetic advance; correlation; path analysis; image analyzer; diversity analysis

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Introduction

Pongamia pinnata (L.) Pierre, synonymously known as *Pongamia glabra* Vent., *Derris indica* (Lam) Bennett., *Millettia novo-guineensis* Kane & Hat. and *Cytisus pinnaus* L. is an arboreal legume, belonging to the subfamily Papilionoideae and specifically the tribe Millettieae. This medium-size tree, being indigenous to the Indian subcontinent and south-east Asia (Malaysia and Indonesia), have been successfully introduced to humid tropical regions of the world as well as parts of Australia, New Zealand, China and the USA (Scott et al. 2008). Historically, this plant has been used in India and neighboring regions as a source of traditional medicines, green manure, timber, fish poison and fuel. The mature tree can withstand water logging and slight frost, and highly tolerant to salinity. *Pongamia* can help in restoration of fertility especially in degraded soils owing to its nitrogen fixing ability. Extracts from the plant are known to have the medicinal properties and effects on a wide array of organisms including insect and pests, molluses and nematodes (Baswa et al. 2001; Latha et al. 2001; Srinivasan et al. 2003). *Pongamia* seed oil resembling ground nut oil (*Arachis hypogaea* L.) with its fatty acid composition and high oleic acid content (45%–70% w.t.) is a source for a number of bioactive compounds including flavonoids and furan-flavonoids, which has medicinal uses for rheumatism, skin diseases, etc. (Parmar et al. 1976; CSIR 1988). More importantly, *P. pinnata* has recently been recognized as a viable source of oil for the burgeoning bio-fuel industry (Karmee and Chadha 2005). Added to this, the low-temperature operability of the corresponding methyl esters is superior to that of Jatropha oil because of the relatively high percentage of oleic acid in karanj oil (Srivastava et al. 2008).

P. pinnata contributes significantly as a source of non-edible oil feedstock and is capable of growing on marginal lands (Hill et al. 2006). However, for meeting the future demands of feedstock for bio-diesel, it is important to establish extensive plantations from elite source. Although it may look promising, it lacks the improved germplasm for large scale plantation. Seed poly-

morphism in *P. pinnata* has been found to play an important role in seed germination, seedling survival and growth (Pathak et al. 1980). The knowledge of genetic variability and correlation between pod and seed traits linked with progeny field performance at early stage and diversity analysis among the collected germplasm is considered to provide necessary information for further genetic improvement of the species in maximizing the yield. The challenging task is to screen the naturally available genetic variation by evaluation of progeny traits for higher productivity. The progenies with better traits, not only have better adaptability to the study site, but also perform better for fruiting and seed oil yield, which consecutively provides opportunity for mass clonal propagation. Considering the present day scenario, an effort has been made to evaluate the extent of genetic variability, correlation among pod and seed traits with progeny traits along with analysis of genetic diversity as scope for further genetic improvement programme.

Materials and methods

Plant materials

An extensive wild germplasm exploration survey was conducted to identify the high yielding Candidate Plus Trees (CPTs) of *P. pinnata* at fruiting stage from different predominant naturalized locations in Jharkhand, India. Since *P. pinnata* was grown as wild and had no definite geometry with neighboring trees for comparison, the selection was made by using single tree selection method based on phenotypic assessment of characters of economic importance viz. yield potential, crown spread, total height, girth at breast height, age of the tree, free from pest and diseases, seed size and seed weight. A total of 24 CPTs (morphologically superior trees), covering a latitude and longitudinal range from N 22° to 24° 50' and E 83° 30' to 87°, were selected (Table 1, Fig 1). Three kilograms of mature pods from each CPT were collected following a random sampling procedure from all the four directions of the crown of each selected tree during fruiting season in April–June, 2005. The observations for 13 quantitative traits (4 pod traits, 6 seed traits of parent trees and 3 progeny traits) were recorded at Forest Research Centre, Institute of Forest Productivity, Mandar, Ranchi district during 2005–2007.

Study site

The area under Forest Research Centre (latitude: 23°27′40″ N, longitude: 85°05′56″ E, and altitude: 2 320 ft, m.s.l. approx.) is a semi-arid type of climate receiving mean annual rainfall of 1231.6 mm with mean number of rainy days for 73.6. Annual minimum and maximum temperature is 17.7°C and 30.2°C, respectively, with lowest temperature in January and highest temperature in May every year. Soils of the study area are characterized by pH (5.7), EC (35 siemens·m^{-1}), Organic carbon (0.33%), Nitrogen (0.0105%), Phosphorus (0.0011% and Potassium (0.0074%).

Table 1. Locational details of *Pongamia pinnata* candidate Plus Trees (CPTs) selected in Jharkhand, India

CPTs	District	Location/Village	Latitude	Longitude	Altitude (m)	Age (years)	Height (m)	DBH (cm)	Seed yield (kg·a^{-1})	Crown area (m^2)
CPT-1	Ranchi	Barhe	23°28′36″N	85°01′06″E	610	75	17	125	200	333.1
CPT-2	Gumla	Indrakela Girijatoli	23°07′02″N	84°33′21″E	520	25	12	50	60	162.8
CPT-3	Lohardaga	Chechra Nawadih	23°26′17″N	84°38′36″E	590	80	14	107	300	194.7
CPT-4	Lohardaga	Kandra	23°21′06″N	84°39′16″E	570	85	10	103	250	297.0
CPT-5	Simdega	Piosokra	22°35′47″N	84°40′49″E	370	55	13.6	92	150	193.5
CPT-6	Lohardaga	Bather nawatana	23°33′02″N	84°54′43″E	640	50	13.7	128	100	312.4
CPT-7	Garhwa	Vishrampur	23°55′30″N	83°46′11″E	410	20	10.3	35	50	150.6
CPT-8	Chatra	Utta sangra	24°14′15″N	85°00′15″E	640	60	15.5	92	250	260.0
CPT-9	Hazaribag	Nawakutar	23°54′19″N	85°19′04″E	610	100	17	114	160	289.4
CPT-10	Hazaribag	Gramurwan	24°27′10″N	85°31′42″E	370	20	11.9	70	35	142.0
CPT-11	Koderma	Bariyadi	24°27′21″N	85°46′12″E	380	40	10.2	93	85	239.0
CPT-12	Ranchi	Chuttupallu	23°27′45″N	85°28′39″E	630	60	11.5	77	100	198.5
CPT-13	Saraikela	Hatnada Tal-tola	22°51′42″N	85°56′55″E	390	20	11	55	40	122.7
CPT-14	Dhalbum	Dhalbumghar	22°27′10″N	86°37′09″E	350	20	8.0	50	45	69.4
CPT-15	Ranchi	Pansakam	23°09′04″N	85°28′40″E	500	80	21	98	150	306.2
CPT-16	Gumla	Hutar	23°16′31″N	85°03′21″E	790	70	14.5	90	120	399.2
CPT-17	Gumla	Bishrampur Jhatnitoli	23°08′22″N	84°46′47″E	800	80	12.7	140	140	331.5
CPT-18	Gumla	Bombibary	22°52′39″N	84°53′36″E	500	50	12.4	105	100	323.5
CPT-19	Chaibasa	Murumbura	22°52′35″N	85°18′15″E	690	80	16	140	200	333.1
CPT-20	Khunti	Itae dartoli	23°03′05″N	85°13′40″E	700	50	18.5	122	100	342.9
CPT-21	Ranchi	Jamun Tolli	23°33′55″N	85°05′05″E	650	60	16	158	140	289.4
CPT-22	Giridih	Bangabad	24°17′11″N	86°21′55″E	390	50	9.9	86	150	281.9
CPT-23	Ranchi	Chund	23°28′40″N	85°10′17″E	790	60	12.0	93	100	229.5
CPT-24	Ranchi	Pandu	23°17′07″N	85°10′35″E	810	65	10.3	102	130	248.7

Pod characters

The pods were cleaned, dried and stored in muslin bags at ambient conditions. All pots were dried under similar temperature and humidity conditions to reach constant weight. A total of 300 healthy pods (hundred in each replication) were randomly selected and observations for four pod traits viz. pod length, pod width, pod thickness and 100-pod weight were measured as mentioned in Table 2.

Seed characters

Samples of 300 seeds were randomly collected from each CPT to make three replications of each 100-seed. Measurement of morphometric traits viz. seed length, seed breadth, aspect ratio and 2D surface area, was done using Image analyzer (Leica Quantimet called QWin 500). Seeds were spread on a glass platform of macro-viewer for each replication and images were captured using charge coupled device (CCD) camera in the software of Quantimet 500 or Qwin (Name of software). The Qwin identifies the object based on our specification for seed colour and calibrates the captured images to actual scale. The various 2-dimensional measurements of the detected images and other parameter were measured as mentioned in Table 2.

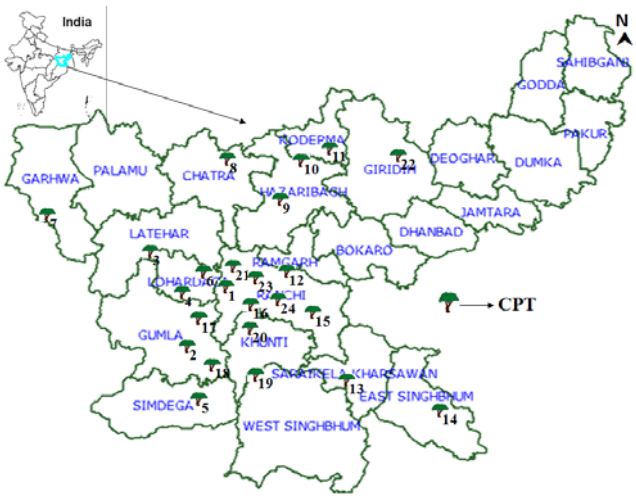

Fig. 1 Distribution map of candidate plus tree (Detail of number representation is in Table 1)

Table 2. Methodology for measuring pod, seed and progeny traits of *Pongamia pinnata*

Sl. No.	Traits	Method
1.	Pod length (mm)	Length of the pod at longest side was measured using vernier caliper, and average value was computed.
2.	Pod width (mm)	Length of the pod at shortest side was measured using vernier caliper, and average value was computed.
3.	Pod thickness (mm)	Thickness of the pod was measured using vernier caliper, and average value was computed.
4.	100 – pod weight (g)	Weight of 100-pods weighed on electronic balance and average value were calculated.
5.	2D surface area (mm^2)	2D surface area of the seed in the direction of measurement.
6.	Seed length (mm)	Length of the seed at longest side.
7.	Seed breadth (mm)	Length of the object at shortest side.
8.	Aspect ratio	Length was divided by breadth.
9.	100 – seed weight (g)	Weight of 100 seeds weighed on electronic balance was measured in grams.
10.	Ratio of pod to seed	100-pod weight was divided by 100-seed weight.
11.	Plant height (cm)	Length of the plant from ground level to tip.
12.	Collar diameter (cm)	Stem diameter near the ground level.
13.	Volume index (cm^3)	[Collar diameter (cm)]^2 × Plant height (cm)] (Manavalan 1990).

The progenies

Seeds of all the CPTs were uniformly pre-treated by soaking in cold water for 24 h. Three hundred pre-treated seeds of each CPT were directly sown in polythene bags of dimension of 30 cm×12 cm×10 cm filled with potting mixture of soil, sand and farmyard manure (2:1:1) in three replicates of each 100-seed during July 2005. Samples of six one-year-old seedlings were planted in the field (pit size 45cm × 45cm × 45cm) in July 2006 in a randomized complete block design with three replications at spacing 3.5 m×3.5 m for field evaluation at Forest Research Centre. At juvenile stage (30 months after sowing (MAS)), observation were recorded on the trial for plant height (m), collar diameter (cm), at periodical bimonthly intervals viz., 2 months after planting (MAP), 4 MAP, 6 MAP, etc. The data recorded at 18 MAP alone were considered for variability, correlation and diversity studies. Volume index was calculated as mentioned in Table 2.

Data analysis

The pod and seed parameters and progeny traits were analyzed using Analysis of variance (ANOVA) and Duncan Multiple Range Test (DMRT) to understand the significant difference among the pod, seed and progeny trait of CPTs under consideration (Gomez and Gomez 1984). The phenotypic variation for each trait was partitioned into components due to genetic (hereditary) and non-genetic (environmental) factors and estimated using the following formula (Johanson et al. 1955)

$$V_p = M_{SG} / r \quad (1)$$

$$V_g = (M_{SG} - M_{SE})/r \quad (2)$$

$$V_e = M_{SE} \quad (3)$$

where, M_{SG}, M_{SE} and r are the mean squares of CPTs, mean squares of error and number of replications, respectively. The phenotypic variance (V_p) is the total variance among phenotypes. The genotypic variance (V_g) is the part of the phenotypic variance that can be attributed to genotypic differences among the phenotypes, and the error variance (V_e) is part of the phenotypic variance due to environmental effects. To compare the variation among traits, phenotypic (P_{CV}) and genotypic (G_{CV}) coefficients of variation were computed according to the method suggested by Burton (1952):

$$P_{CV} = (\sqrt{V_p}/X) \times 100 \quad (3)$$

$$G_{CV} = (\sqrt{V_g}/X) \times 100 \quad (4)$$

where, V_p, V_g and X are the phenotypic variance, genotypic variance and grand mean for each pod and seed-related trait, respectively. Broad sense heritability (h^2B) was calculated according to Allard (1999) as the ratio of the genotypic variance (V_g) to the phenotypic variance (V_p). Genetic advance (GA) was estimated in accordance with Johanson et al. (1955) as follows:

$$G_A = K \cdot h^2B \cdot \sqrt{V_p} \quad (5)$$

$$G_A = (G_A/X) \times 100 \quad (6)$$

where, K is the selection differential (2.06 for selecting 5% of the genotypes); G_A is as % of the mean. Phenotypic (r_p) and genotypic (r_g) correlations were further computed to examine inter-character relationships among seed and seedling traits following Goulden (1952) as:

$$r_p = C_{ovp}(x_1, x_2)/[V_p(x_1) \cdot V_p(x_2)]^{1/2} \quad (7)$$

$$r_g = C_{ovg}(x_1, x_2)/[V_g(x_1) \cdot V_g(x_2)]^{1/2} \quad (8)$$

where, C_{ovp} and C_{ovg} are phenotypic and genotypic covariances for any two traits x_1 and x_2, respectively, and Vp and Vg are the respective phenotypic and genotypic variances for those traits. Path coefficient analysis was done using genotypic correlation coefficients following Dewey and Lu (1959). The genetic diversity was calculated by using non-hierarchical Euclidian cluster analysis (Spark 1973).

Results

Analysis of variance for pod, seed and progeny traits reveled that there was significant variation among CPTs (Table 3).

Table 3. Mean performance of selected genotypes for pod, seed and progeny traits in *Pongamia pinnata*

CPTs	Pod traits					Seed traits					Progeny traits (30 MAS)		
	Length (mm)	Width (mm)	Thickness (mm)	100-Pod weight (g)	2D area (mm^2)	Length (mm)	Breadth (mm)	Aspect ratio	100-seed weight (g)	Pod/seed ratio	Height (cm)	Collar diameter (cm)	Volume index (cm^3)
CPT-1	51.3^{d}	26.3^{a}	9.6^{k}	231.0^{m}	278.4^{fghi}	21.8^{h}	15.8^{d}	1.4^{j}	115.0^{n}	2.1^{jklm}	95.3^{f}	1.6^{d}	261.0^{c}
CPT-2	45.1^{i}	18.9^{h}	11.8^{bcde}	254.2^{l}	235.5^{l}	24.8^{cde}	12.8^{h}	2.0^{a}	106.1^{o}	2.4^{de}	96.5^{f}	1.6^{d}	274.5^{c}
CPT-3	55.9^{c}	20.4^{fg}	12.0^{bcd}	358.1^{ef}	284.6^{efg}	23.4^{efg}	15.2^{e}	1.5^{fghi}	123.0^{mn}	2.9^{a}	140.2^{abcd}	3.2^{a}	1489.5^{ab}
CPT-4	51.0^{de}	26.6^{a}	11.5^{defg}	407.5^{d}	346.1^{ab}	25.6^{bc}	17.6^{a}	1.5^{hij}	171.5^{de}	2.4^{def}	147.0^{abc}	2.8^{abc}	1166.4^{abcde}
CPT-5	50.0^{defg}	20.3^{fg}	12.0^{bcd}	356.4^{ef}	326.1^{bc}	24.1^{def}	16.6^{bc}	1.5^{il}	161.5^{fg}	2.2^{fghi}	128.8^{bcde}	2.4^{abcd}	$837.2^{abcdefg}$
CPT-6	58.2^{b}	25.0^{bc}	12.7^{a}	474.4^{a}	344.6^{ab}	26.6^{b}	17.5^{a}	1.5^{fghi}	165.8^{ef}	2.9^{a}	125.7^{bcde}	2.4^{abcd}	698.9^{cdefg}
CPT-7	56.7^{bc}	26.4^{a}	11.5^{defg}	357.8^{ef}	314.6^{cd}	24.5^{cde}	16.9^{b}	1.5^{hij}	151.5^{hij}	2.4^{def}	125.5^{bcde}	2.7^{abc}	$929.9^{abcdefg}$
CPT-8	47.7^{gh}	19.8^{gh}	12.3^{ab}	284.6^{jk}	286.8^{efg}	23.5^{defg}	16.2^{c}	1.5^{hij}	135.3^{l}	2.1^{hijk}	146.5^{abc}	2.9^{ab}	1230.0^{abcd}
CPT-9	50.0^{defg}	23.7^{de}	12.0^{bcd}	352.4^{efg}	315.8^{cd}	24.1^{def}	17.3^{a}	1.4^{j}	154.0^{ghi}	2.3^{efg}	135.2^{bcde}	2.9^{ab}	1128.2^{abcdef}
CPT-10	65.7^{a}	23.6^{de}	11.0^{ghi}	451.8^{c}	332.3^{abc}	26.8^{ab}	15.2^{e}	1.8^{bc}	174.8^{d}	2.6^{bc}	149.0^{ab}	3.2^{a}	1486.8^{ab}
CPT-11	57.7^{bc}	23.1^{e}	12.1^{bc}	358.9^{ef}	297.4^{def}	25.6^{bc}	14.4^{g}	1.8^{b}	183.7^{bc}	2.0^{klmn}	145.2^{abc}	3.0^{ab}	1352.3^{abc}
CPT-12	58.5^{b}	26.0^{ab}	10.5^{ij}	337.3^{gh}	243.7^{kl}	24.9^{cd}	15.1^{e}	1.7^{d}	156.1^{gh}	2.2^{ghij}	126.2^{bcde}	2.6^{abc}	$882.2^{abcdefg}$
CPT-13	50.3^{def}	23.4^{de}	9.7^{k}	274.8^{k}	270.9^{ghij}	24.0^{def}	14.6^{fg}	1.6^{de}	128.9^{lm}	2.1^{ghij}	112.0^{ef}	1.9^{cd}	445.9^{fg}
CPT-14	51.5^{d}	24.9^{bc}	11.7^{bcde}	423.1^{d}	305.5^{de}	24.4^{cde}	16.6^{bc}	1.5^{hij}	176.9^{cd}	2.4^{def}	146.0^{abc}	3.1^{ab}	1412.8^{ab}
CPT-15	48.5^{fgh}	21.3^{f}	11.8^{bcde}	303.1^{i}	281.8^{fgh}	23.7^{def}	15.2^{e}	1.6^{efgh}	135.7^{l}	2.2^{efghj}	125.2^{bcde}	1.9^{cd}	482.4^{efg}
CPT-16	45.1^{i}	18.7^{h}	11.0^{fgh}	257.6^{l}	256.5^{jk}	23.6^{def}	14.4^{g}	1.6^{de}	121.6^{mn}	2.1^{ghij}	134.3^{bcde}	2.6^{abc}	973.9^{abcdef}
CPT-17	48.8^{efgh}	27.1^{a}	10.1^{jk}	329.2^{h}	287.8^{efg}	22.2^{gh}	17.5^{a}	1.3^{k}	148.5^{hij}	2.2^{efgh}	150.3^{ab}	3.2^{a}	1518.7^{a}
CPT-18	47.4^{h}	21.1^{f}	11.6^{cdef}	366.5^{e}	290.3^{efg}	25.6^{bc}	14.6^{g}	1.8^{bc}	137.2^{kl}	2.7^{b}	122.5^{cde}	2.5^{abc}	795.3^{bcdefg}
CPT-19	65.6^{a}	23.7^{de}	11.7^{cde}	542.4^{a}	351.2^{a}	27.9^{a}	17.4^{a}	1.6^{def}	217.9^{a}	2.5^{cd}	164.3^{a}	2.9^{ab}	1435.8^{ab}
CPT-20	43.2^{i}	20.4^{fg}	10.7^{hi}	333.3^{h}	258.4^{ijk}	23.7^{def}	14.2^{g}	1.7^{d}	185.0^{b}	1.8^{n}	141.5^{abc}	3.0^{ab}	1298.3^{abcd}
CPT-21	48.2^{fgh}	22.8^{e}	10.7^{hi}	343.5^{fgh}	284.7^{efg}	24.3^{cde}	14.5^{g}	1.8^{cd}	144.7^{jk}	2.4^{def}	136.8^{bcd}	2.7^{abc}	1049.4^{abcdef}
CPT-22	44.7^{i}	18.7^{h}	10.0^{jk}	233.3^{m}	273.1^{ghij}	23.6^{def}	15.0^{ef}	1.6^{defg}	125.9^{m}	1.9^{mn}	123.0^{cde}	2.3^{abcd}	703.4^{cdefg}
CPT-23	44.4^{i}	24.3^{cd}	11.3^{efg}	296.7^{ij}	270.3^{ghij}	22.7^{fgh}	15.3^{e}	1.5^{ghi}	144.7^{jk}	2.1^{ijkl}	116.2^{def}	2.2^{bcd}	629.9^{defg}
CPT-24	49.1^{defgh}	23.2^{de}	9.8^{k}	276.3^{k}	260.6^{hijk}	20.3^{i}	16.2^{c}	1.3^{k}	146.1^{ij}	1.9^{lmn}	148.7^{ab}	3.0^{ab}	1386.1^{abc}
Mean	51.4	22.9	11.2	341.8	291.5	24.2	15.7	1.6	150.5	2.3	132.6	2.6	994.5
SEM	0.7	0.4	0.2	5.7	6.6	0.4	0.1	0.03	2.7	0.1	7.3	0.3	206.0
CD 5%	2.1	1.1	0.5	16.7	19.2	1.2	0.4	0.08	7.8	0.2	21.3	0.7	599.5

Notes: Traits by the same superscript letter are not significantly different at $p = 0.05$. MAS is Months after sowing.

Variability of CPT-19 had maximum for seven traits viz, pod length (65.6 mm), 100-pod weight (542.4 g), 2D surface area (351.2 mm^2), seed length (27.9 mm), seed breadth (17.4 mm), 100-seed weight (217.9 g) and plant height (164.3 cm). However, maximum volume index was recorded in CPT-17 (1 518.8 cm^3), followed by CPT-3 (1 489.5 cm^3), CPT-10 (1 486.8 cm^3), CPT-19 (1 435.8 cm^3) and CPT-14 (1 412.8 cm^3). Lowest 100-pod and 100-seed weight were recorded in CPT-1 (231.0 g) and CPT-2 (106.1 g) respectively.

The genetic estimates of pod, seed and progeny growth performance are shown in Table 4. There were fair difference between genotypic coefficients of variation and phenotypic coefficients of variation for all traits except progeny traits. All the pod and seed traits showed high heritability and progeny growth traits had moderate heritability. The 100-pod weight exhibited highest heritability (more than 98.4%) followed by 100-seed weight (96.9%). The 100-pod weight and 100-seed weight expressed high heritability (98.4%, 96.9%), accompanied with high genetic advance (46.0%, 34.9%). Volume index expressed moderate heritability (47.4%), accompanied with high genetic advance (48.4%).

In general, the genotypic correlation coefficient values were higher than corresponding phenotypic values (Table 5). Correlation study of thirteen quantitative traits (4 pod traits, 6 seed traits and 3 progeny traits) revealed that among 156 (78 genotypic and 78 phenotypic) correlations, 31 genotypic and 23 phenotypic combinations were significant at 1% level along with 11 genotypic and 12 phenotypic combinations significant at 5% level. The trait and 100-pod weight expressed positive significant correlation at both genotypic and phenotypic levels with plant height (rg= 0.66, r_p= 0.51), collar diameter (0.59, 0.40) and volume index (0.60, 0.41) at 30 MAS (Months after sowing) respectively. However, pod length (0.43), seed 2D surface area (0.40) and seed breadth (0.42) expressed positive significant correlation only at genotypic level with volume index at 30 MAS. Path analysis of pod, seed and progeny growth traits was carried out to unlock the direct and indirect contributions of pod, seed and progeny growth characters on volume index at 30 MAS. The seed breadth had the highest direct (2.18) and indirect effect on volume index through pod width (1.32) and 2D surface area (1.62). Though 100-seed weight was highly significantly correlated with volume index at 30 MAS, the direct effects were less.

Table 4. Genetic estimates of pod, seed and progeny traits in *Pongamia pinnata*

	Traits	Genotypic coefficient of Variation	Phenotypic coefficient of Variation	Heritability (%)	GA (%) of mean
Pod traits	Length (mm)	12.1	12.3	96.2	24.4
	Width (mm)	11.5	11.8	94.6	22.9
	Thickness (mm)	7.8	8.2	89.4	15.2
	100-pod weight (g)	22.5	22.7	98.4	46.0
Seed traits	2D area (mm^2)	10.8	11.5	88.4	21.0
	Length (mm)	6.6	7.2	82.6	12.3
	Breadth (mm)	8.2	8.4	96.8	16.7
	Aspect ratio	10.4	11.0	90.4	20.4
	100 -seed weight (g)	17.2	17.5	96.9	34.9
	Pod – seed ratio	12.7	13.4	89.9	24.7
Progeny traits *	Plant height (cm)	11.5	14.9	58.9	18.1
	Collar diameter (cm)	15.9	23.3	46.7	22.4
	Volume index (cm^3)	34.1	49.5	47.4	48.4

Note: * shows progeny traits (30 months after sowing).

Table 5. Genotypic (G) and phenotypic (P) correlation coefficient matrix of pod, seed and progeny traits in *Pongamia pinnata*

Traits		Pod width	Pod thickness	100-Pod weight	Seed 2D area	Seed length	Seed breadth	Aspect ratio	100-seed weight	Pod–Seed ratio	Plant Height	Collar Diameter	Volume index
Pod length	G	0.44*	0.22	0.74**	0.61**	0.64**	0.36	.09	0.57**	0.49*	0.43*	0.38	0.43*
	P	0.45*	0.21	0.73**	0.58**	0.61**	0.35	.11	0.55**	0.48*	0.32	0.27	0.29
Pod width	G		-0.19	0.37	0.39	0.05	0.61**	).46*	0.36	0.11	0.11	0.13	0.15
	P		-0.18	0.37	0.37	0.07	0.58**	).41*	0.35	0.12	0.11	0.12	0.12
Pod thickness	G			0.51**	0.49*	0.54**	0.20	.19	0.29	0.55**	0.21	0.21	0.16
	P			0.49*	0.45*	0.50*	0.18	.20	0.27	0.51**	0.20	0.15	0.13
100-Pod weight	G				0.83**	0.79**	0.52**	.07	0.82**	0.63**	0.66**	0.59**	0.60**
	P				0.77**	0.73**	0.50**	.08	0.81**	0.62**	0.51**	0.40*	0.41*
Seed 2D area	G					0.61**	0.74**	).24	0.65**	0.52**	0.53**	0.38	0.40*
	P					0.58**	0.71**	).18	0.60**	0.46*	0.38	0.26	0.28
Seed length	G						0.08	.56**	0.59**	0.57**	0.24	0.09	0.09
	P						0.07	.61**	0.53**	0.51**	0.22	0.18	0.16
Seed breadth	G							).78**	0.45*	0.23	0.51**	0.41*	0.42*
	P							).75**	0.45*	0.20	0.38	0.28	0.29
Aspect ratio	G								-0.01	0.18	-0.28	-0.29	-0.30
	P								-0.01	0.19	-0.17	-0.11	-0.12
100-seed weight	G									0.09	0.78**	0.67**	0.69**
	P									0.04	0.60**	0.47*	0.49*
Pod–Seed ratio	G										0.11	0.18	0.16
	P										0.08	0.11	0.09
Plant Height	G											0.96**	0.97**
	P											0.84**	0.89**
Collar Diameter	G												0.99**
	P												0.97**

Notes: *significant at $p = 0.05$, **significant at $p = 0.01$

Twenty-four accessions of *P. pinnata* were placed under six clusters on the basis of non-hierarchical Euclidian cluster analysis (Table 6). The maximum numbers of accessions (seven) were grouped in cluster IV, followed by cluster III with five accessions. Whereas cluster II and I had two and four accessions, cluster V, VI had three accessions respectively. In the present study, the clustering pattern of the genotypes indicated that geographical diversity may not be related to genetic diversity. This was proved by tendency of genotypes from diverse eco-geographic regions to group together in the same cluster or scattered distribution of genotypes of same geographic origin in different clusters. Intra- and inter-cluster distance ranged from 26.2 to 114.0 and 146.2 to 1201.2 respectively. Intra-cluster distance was maximum in cluster IV (114.0) with 7 accessions and minimum in cluster II (26.2) with 2 accessions respectively (Fig. 2). Highest Inter-cluster distance was between cluster V and II (1201.2), followed by cluster II and I (1177.4), suggesting that there is wide genetic diversity between these groups. The minimum inter-cluster distance was between cluster I and V (146.2).

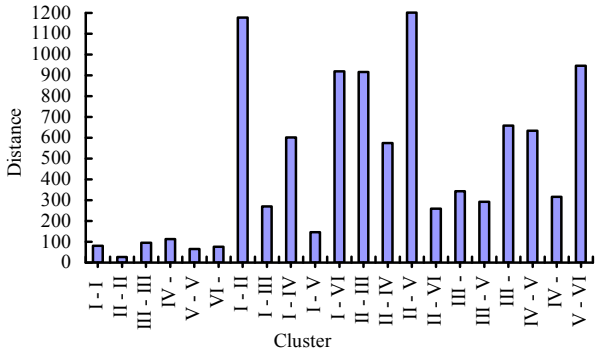

Fig. 2 Estimates of inter- and intra-cluster distances for pod, seed and progeny traits in *P. pinnata* accessions

Cluster means expressed significant variation among clusters for all the traits, particularly for volume index. In general, the cluster I and cluster II had highest and lowest mean values for most of the traits respectively. Cluster V had maximum for trait volume index (Table 7).

Discussion

The study on pod and seed morphometric traits of the wild genotypes of *P. pinnata* is often considered to be the first, useful and easy step in ascertaining the genetic variability of the populations. Significant variation in 100-pod and 100-seed weight depends on reserve food material, which is produced as a result of double fertilization (endosperm) and is dominated by the maternal traits along with influence of the nutrient availability at the time of seed setting and prevailing environmental factors (Allen 1960; Johnsen et al. 1989). Hence, in the present study, various CPTs exhibiting significant variability in pod and seed traits could be attributed to fact that the species grows over a wide range of rainfall, temperature and soil type, indicating the marked difference in selection pressure. Habitat influences on pod and seed traits have also been reported in number of tree species like *Jatropha curcas* (Ginwal et al. 2004; Rao et al. 2008; Kaushik et al. 2007a), *Madhuca latifolia* (Divakara and Krishnamurthy 2009) and *Pongamia pinnata* (Kaushik et al. 2007b).

Table 6. Composition of Euclidean cluster for pod, seed and progeny traits in *P. pinnata* accessions

Clusters	Number of accessions	Accessions (CPTs)
I	4	CPT-10, CPT-11, CPT-14, CPT-19
II	2	CPT-1, CPT-2
III	5	CPT-4, CPT-8, CPT - 9, CPT-20, CPT-21
IV	7	CPT-5, CPT-6, CPT-7, CPT-12, CPT-16, CPT-18, CPT-22
V	3	CPT-3, CPT-17, CPT-24
VI	3	CPT-13, CPT-15, CPT-23

Table 7. Cluster mean value for pod, seed and progeny traits in *P. pinnata* accessions

Clusters	Pod length (mm)	Pod width (mm)	Pod thickness (mm)	100-pod weight (g)	Seed 2D Area (mm^2)	Seed length (mm)	Seed breadth (mm)	Aspect ratio (mm)	100 seed weight (g)	Pod-seed ratio	Plant height (cm)	Collar diameter (cm)	Volume index (cm^3)
I	60.1	23.8	11.6	444.0	321.6	26.2	15.9	1.7	188.4	2.4	151.1	3.1	1422.0
II	48.2	22.6	10.7	242.6	257.0	23.3	14.3	1.7	110.6	2.2	95.9	1.6	267.7
III	48.0	22.7	11.4	344.2	298.4	24.2	16.0	1.5	158.1	2.2	141.4	2.9	1174.4
IV	51.5	22.3	11.3	340.5	292.7	24.7	15.7	1.6	145.7	2.3	126.6	2.5	831.5
V	51.3	23.5	10.6	321.2	277.7	22.0	16.3	1.4	139.2	2.4	146.4	3.1	1464.8
VI	47.8	23.0	10.9	291.5	274.3	23.5	15.0	1.6	136.5	2.1	117.8	2.0	519.4

Though the selection of superior trees is carried out intensively and clonal superiority plants are established, genetic superiority needs to be determined. The genetic estimates can be very useful tools in predicting the amount of gain expected in short period. The variation among genotypes is commonly used as an estimate of total genetic variation to calculate the degree of genetic control for a particular trait (Foster & Shaw 1988). Marginal difference between PCV (phenotypic coefficients of variation) and GCV (genotypic coefficients of variation) and high estimates of heritability (broad sense) for all pod and seed traits

revealed the heritable nature of variability present. Relatively high value of genotypic variance resulted in high estimates of heritability, contributing to the high genetic gains in thirteen quantitative traits (4 pod traits, 6 seed traits and 3 progeny traits). Gains from tree breeding programs depend on the type and extent of genetic variability. In the present study, the genotypic coefficients of variation and the genetic gain were found to be comparatively higher for important traits such as volume index, 100-pod and 100-seed weight. The trait for volume index may be changed considerably by selecting the superior 5% of the genotypes. High heritabilities accompanied by high genetic advance for growth parameters have been reported in other tree species like *Jatropha curcas* (Ginwal et al. 2004; Rao et al. 2008), indicating possibility of genetic improvement in growth parameters.

The ultimate goal of the tree improvement is to improve growth and yield traits of tree species. Growth and yield traits are complex and the product depends on the interplay of many physiological and morphological attributes, hence improvement based on performance of tree species alone might prove to have less effective. In genetic improvement of growth and yield traits of *P. Pinnata*, clear understanding of the relationships among different pod, seed and growth traits is very essential. As variation among clones is used for estimation of genetic variation and genetic gain, co-variance estimates between traits can be used to estimate genetic correlations between the traits (Foster 1986). Correlation shows the extent of association between seed traits, which may form additional criteria for selection in breeding program. Correlated quantitative traits are of a major interest in an improvement program, as the improvement of one character may cause simultaneous correlated changes in the other characters. Genotypic and phenotypic correlation coefficients between various characters revealed that magnitude of correlation coefficient at genotypic level was higher than their corresponding phenotypic coefficient of correlations. The genotypic correlation is an estimated value, whereas, phenotypic correlation is a derived value from the genotype and environmental interaction (Chaturvedi and Pandey 2004). The genotypic correlation indicates genotypic association among the traits and is, therefore, a more reliable estimate value for examining the degree of relationship between character pairs.

Path analysis of pod, seed and progeny growth traits revealed that, even though seed breadth has slightly low (0.42) correlation coefficients, it has highest direct (2.18) and indirect effect on volume index through pod width (1.32) and 2D surface area (1.62). Hence seeds with good breadth may be selected for producing better progenies in addition to 100-pod and seed weight. Though 100-seed weight is highly significantly correlated with volume index at 30 MAS, the direct effect is less. Positive correlation between seed weight and seedling height was found in *Pinus* spp. but it disappeared with the growing age of the seedlings (Righter 1945). However correlation between seed weight and plant height was observed in *Pinus taeda* till 15 years (Robinson and Van Buijtenen 1979). Khalil (1981) stated that, 1000-seed weight and plant height in *Picea glauca* at four years appeared significant positive correlation. Hence, seed weight may be an index among the criteria for selection of plus trees. Among different seed traits, viz., seed coat, gametophyte and embryo weight, the embryo weight had strong relation with seedling growth traits in *Pinus elliotti* (Surles et al. 1993). In Douglas fir, similar contribution of seed weight to seedling height was reported (Sorenson and Campbell 1993).

Genetic diversity in plant species is a gift to mankind as it forms the basis for selection and further improvement of species. The information on the genetic structure and diversity relationship of CPT provides a basis for planning and conducting future collections and efficient utilization of genetic resources to realize the potentiality for maximizing growth and yield. Various statistical tools like Mahalanobis D^2 analysis, cononical and principal component analysis are helpful in deriving genetic information from quantitative data. The D^2 statistic is one of the powerful tools to assess the relative contribution of different component traits in the total diversity and to quantify the degree of divergence between populations and to choose genetically diverse parents for obtaining desirable recombination.

The clustering pattern in this study revealed that geographical diversity was not necessary to be related to genetic diversity. This kind of genetic diversity might be due to differential adoption methods, selection criteria, natural selection pressure and environment factors (Vivekananda and Subramanian 1993). This indicated that genetic drift produced greater diversity than the geographic diversity (Singh et al. 1996). Absence of any relationships between genetic diversity and geographical distribution is in accordance with the findings of Kaushik et al. (2007a) and Gohil and Pandya (2008) in *Jatropha curcas.* The trees that originated in one region were distributed into different clusters, indicating that trees with same geographic origin could have undergone changes for different characters under selection.

Cluster IV (114.01) and cluster III (95.50) showed maximum intra-cluster distances. Hence genotypes in cluster IV (CPT-5, CPT-6, CPT-7, CPT-12, CPT-16, CPT-18, CPT-22) and cluster III (CPT-4, CPT-8, CPT-9, CPT-20, CPT-21) were most heterogeneous and can be best used within group hybridization. Maximum inter-cluster distance (1201.23) was between cluster V and II, followed by cluster II and I (1177.40), indicating that there was wider genetic diversity between the trees in these groups. Since, wide diversity exists between the cluster V and II, followed by cluster II and I, the crosses between CPTs of these clusters may result in substantial segregate for thirteen quantitative traits (4 pod traits, 6 seed traits and 3 progeny traits) and help in further selection for overall improvement of species. This kind of study can help to identify the better genotypes of *P. pinnata* having better yield and oil content.

From the above study, the traits like 100-pod and seed weight were highly correlated with growth traits of tree. In addition, pod length, 2D surface area and seed breadth expressed correlation with volume index at 30 MAS. Hence identification of good CPTs may be advantageous based on seed weight, size and shape traits. Since traits viz. 100-pod weight, 100-seed weight and volume index have high heritability and genetic advance, these traits may be considered for further improvement by breeding. CPT-19 is found to be superior on the basis of these traits, viz., 100-pod weight, 100-seed weight, seed breadth and volume in-

dex, hence seeds of these CPT may be importance for massive afforestation programme. The present study can however serve as a pointer at later stages of study especially on seed and oil yield.

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References

Allard RW. 1999. *Principles of Plant Breeding* (2nd edt). New York: John Wiley & Sons, p. 254.

Allen GS. 1960. Factors affecting the viability and germination behaviour of coniferous seed. IV. Stratification period and incubation temperature, *Pseudostuga menziesii* (Mirb.) Franco. *For Chron*, **36**: 18–19.

Baswa M, Rath CC, Dash SK, Mishra RK. 2001. Antibacterial activity of karanja (*Pongamia pinnata*) and neem (*Azadirachta indica*) seed oil: a preliminary report. *Microbios*, **105**: 183−189.

Burton GW. 1952. Quantitative inheritance in grasses. Proc. 6th Intl. Grassland Cong. 1: 227–283. (6th ed.) Iowa, USA: Iowa State Univ. Press, p. 267.

Chaturvedi OP, Pandey N. 2004. Correlation and path analysis studies between biomass and other characters in *Bombax ceiba* L. *Silvae Genet*, **53**: 5−6.

CSIR. 1988. *The Wealth of India—Raw Materials* (vol. 3). New Delhi: Council of Scientific and Industrial Research Publication, pp. 206–211.

Dewey DR, Lu KH. 1959. A correlation and path coefficient analysis of components of crested wheat grass seed production. *Agron J*, **51**: 515−518.

Divakara BN, Krishnamurthy R. 2009. Genetic variability, association and divergence studies in seed traits and oil content of *Madhuca latifolia* Macb. accessions. *J Oilseeds Res*, **26**: 686−689.

Foster GS, Shaw DV. 1988. Using clonal replicates to explore genetic variation in a perennial plant species. *Theor Appl Genet*, **76**: 788−794.

Foster GS. 1986. Provenance variation of eastern cottonwood in the lower Mississippi valley. *Silvae Genet*, 35: 32-38.

Ginwal HS, Rawat PS, Srivastava RL. 2004. Seed source variation in growth performance and oil yield of Jatropha curcas Linn. in Central India. *Silvae Genet*, **53**: 186–92.

Gohil RH, Pandya JB. 2008. Genetic diversity assessment in physic nut (*Jatropha curcas* L.). *International Journal of Plant Production*, **2**(4): 321−326.

Gomez AK, Gomez AA. 1984. *Statistical procedure for agricultural research*. John Wiley and Sons, Inc., p. 698.

Goulden CH. 1952. Some distance properties of latent root and vector methods used in multivariate analysis. *Biometrica*, **53**: 325−338.

Hill J, Nelson E, Tilman D. 2006. Environmental, economic and energetic costs and benefits of biodiesel and ethanol biofuels. *Proc Natl Acad Sci*, **103**: 11206−11210.

Johanson HW, Robinson HF, Comstock RE. 1955. Estimate of Genetic and environmental variability in soyabeans. *Agron J*, **47**: 314−318.

Johnsen O, Dietrichson J, Skaret G. 1989. Phenotypic changes in progenies of northern clones of *Picea abies* (L.) Karst. grown in a southern seed orchard. III. Climate changes and growth in a progeny trial. *Scand J For Res*, **4**: 343–350.

Karmee SJ, Chadha A. 2005. Preparation of biodiesel from crude oil of *Pongamia pinnata*. *Bioresour Technol*, **96**: 1425–1429.

Kaushik N, Kumar K, Kumar S, Kaushik N, Roy S. 2007a. Genetic variability and divergence studies in seed traits and oil content of Jatropha (*Jatropha curcas* L.) accessions. *Biomass Bioenergy*, **31**: 497–502.

Kaushik N, Kumar S, Kumar K, Beniwal RS, Kaushik N, Roy S. 2007b. Genetic variability and association studies in pod and seed traits of *Pongamia pinnata* (L.) Pierre in Haryana, India. *Genet Resour Crop Evol*, **54**: 1827−1832.

Khalil MAK. 1981. Correlation of juvenile height growth with cone morphology and seed weight in white spruce. *Silvae Genetica*, **30**(6): 179−181.

Latha S, Mariamma J, Daniel M. 2001. Studies on the effect of leaf leachates of *Pongamia pinnata* on certain crops and weeds and the soil microflora. *Natl. Acad. Sci Lett*, **24**: 63–68.

Manavalan A. 1990. Seedling vigour and bioproductivity in woody biomass species. Ph.D. Thesis. Madurai: Madurai Kamaraj University.

Parmar BS, Sahrawat KL, Mukerje SK. 1976. *Pongamia glabra*: constituents and uses. *J Sci Ind Res*, **35**: 608–611.

Pathak SK, Gupta K, Debroy R. 1980. Studies on seed polymorphism, germination and seedling growth of Pongamia pinnata. *Indian J For*, **2**: 64–67.

Rao GR, Korwar GR, Shanker AK, Ramakrishna YS. 2008. Genetic associations, variability and diversity in seed characters, growth, reproductive phenology and yield in Jatropha curcas (L.) accessions. *Trees Structure and Function*, **22**: (3): 697–709.

Righter FI. 1945. Pinus: the relationship of seed size and seedling size to inherent vigor. *J. Forest*., **43**: 131−137.

Robinson JF, Van Buijtenen JP. 1979. Correlation of seed weight and nursery bed traits with 5, 10 and 15 year volume in loblolly pine progeny test. *Forest Science*, **35**(4): 591−596.

Scott PT, Pregelj L, Chen N, Hadler JS, Djordjevic MA, Gresshoff PM. 2008. *Pongamia pinnata*: An untapped resource for the biofuels industry of the future. *Bioenerg Res*, **1**: 2−11.

Singh AK, Singh SB, Singh SM. 1996. Genetic divergence in scented and fine genotypes of rice (*Oryza sativa* L.). *Ann Agric Res*, **17**: 163−166.

Sorenson FC, Campbell RK. 1993. Seed weight-seedling size correlation in Douglas-fir: Genetic and environmental components. *Can J of For Res,* **23**(2): 275-285.

Spark DN. 1973. Euclidean cluster analysis algorithm. *Applied Stats*, **22**: 126−130.

Srinivasan K, Muruganandan S, Lal J, Chandra S, Tandan SK, Raviprakash V, Kumar D. 2003. Antinociceptive and antipyretic activities of *Pongamia pinnata* leaves. *Phyto Res*, **17**: 259–264.

Srivastava PK, Verma M. 2008. Methyl ester of karanja oil as an alternative renewable source energy. *Fuel*, **87**: 1673−1677.

Surles SE, White TL, Hodge GR., Duryea ML. 1993. Relationship among seed weight components, seedling growth traits and predicted field breeding values in slash pine. *Canadian Journal of Forest Research*, **239**(8): 1550−1556.

Vivekananda P, Subramaninan S. 1993. Genetic divergence in rainfed rice. *Oryza*, **39**: 60−62.